THE FORMATION OF PROGLOTTIDS IN CROSSOBOTHRIUM LACINIATUM (LINTON).

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I. Introduction and Summary.

The statement that all cestodes form their proglottids by the insertion of each new one between the scolex and the one last formed, has been current in our text-books for years and so far as I know there are no statements to indicate that any other mode of origin ever occurs. Upon turning to the original literature, one finds that the above statement rests upon the descriptions given about 1850 by Leuckart for the genus *Tænia* and closely related forms. This account was confirmed and supplemented by other authors about the same time, but in recent years we only find the process referred to as something established beyond all question as the universal method by which proglottids originate. No recent work so far as I am aware attempts to call this dogma in question, nor do writers longer think it worth while to record the fact that they have confirmed so well established a phenomenon.

Despite this universal acceptance our current description appears to be based upon published accounts which deal with a limited number of forms, though the fact that these accounts have not been contradicted leads one to believe that they describe the process correctly in those species which have been most frequently the subject of investigation.

The cestode, *Crossobothrium laciniatum*, a species occurring as an adult in the "sand shark," *Carcharias littoralis*, of our North Atlantic coast, does not conform to the current account in its method of forming proglottids. In this cestode there first appear from behind forward a considerable number of proglottids (Figs. 1, 2 and 10 of Pl. IV.). These "posterior" segments, as I shall call them, continue to form after the accepted fashion until about 35 of them have been produced. They are marked off by straight transverse lines and not until a considerably later period

do even the oldest of them begin to show the sculpturing of the posterior margin which is so characteristic for the adult.

When these posterior proglottids are about 35 in number, there begin to appear, in the unsegmented region just back of the scolex, others which may be termed the "anterior proglottids." These are produced in the reverse order, *i. e.*, the youngest is the most posterior one.

From such a condition as this (Fig. 10, Pl. IV.) the formation of proglottids continues so that from this time on we may say that the body is segmenting from both ends toward the middle. This process gradually obliterates the unsegmented zone (Fig. 12, Pl. V.) but specimens several centimeters in length may still show such a "zone" at a point near the line dividing the anterior and middle thirds of the body.

When this zone finally becomes obliterated there have been formed in the neighborhood of 50 "anterior" proglottids and upwards of 400 "posterior" ones. With this obliteration of the zone we reach what may be termed the fully formed adult condition as is shown by Fig. 14, *a–n*. *After such a stage is reached no more proglottids are formed.* and the specimen remains unchanged save for the continual maturing and separating off from the posterior end of the motile proglottids. This latter process finally reduces the total number of the segments to such an extent that the condition is perceptible at first glance. Such a specimen which had only 96 proglottids is shown in Fig. 16, *a* and *b*.

When the number of units in the strobilla has been reduced somewhat further, the region between the scolex and the first proglottid begins an active growth and elongates into a neck region as is shown by Figs. 21 and 15. After further elongation and the continual separation of the terminal proglottids, this neck region begins to form segments while there are still some of the most anterior segments of the old chain remaining (Figs. 17 and 24). This new segmentation begins as does that previously described by the appearance of posterior segments and later of anterior ones, and although I have no stages beyond the point shown in Fig. 24, I think one is justified in the opinion that this segmentation is the beginning of a new chain and that soon a new strobilla will be formed which would be with difficulty, if at

all, distinguishable from the one proceeding from the first segmentation. The specimen represented by Fig. 19, Pl. VI., is an exception to the above course of development, in that it shows all the characteristics of a specimen having the non-segmented zone of the early proglottid formation and at the same time an elongated neck. A possible explanation of this as a specimen undergoing regeneration after mutilation is given on p. 215 of this paper.

A discussion of the bearing these facts have upon our views regarding the nature of the cestode body in given on p. 217.

II. Discussion of the Literature.

In my examination of the literature I have relied upon Braun's monograph (Bronn's Thierreich) as a trustworthy summary of the facts established regarding the Cestoda down to about 1896, when the parts of this volume referring to the points in question appeared. Upon the particular subject of the method of proglottid formation the statements of this author are explicit in several instances and agree with the account which has long been current in our text-books. For example, in the introductory section on Cestoda (p. 1167), he says: "besitzen dem nach die Cestoden Zweierlei Theile: Scolex and Proglottiden: letzterer sind in einer Reihe dem Alter nacht geordnet, so das die jüngsten dem scolex am nächsten, die ältesten von diesem am weitesten entfernt sind." On p. 1224, under the paragraph entitled "Intercalation und Verwachsung von Proglottiden," Braun says: "wir haben allen Grund zu der Annahme dass die Proglottiden die ein Bandwurm besitzt oder jemals besessen hat, alle nach einander und zwar in der durch die Stellung gegebenen Reichenfolge am Scolex sich gibildet haben, dass also niemals durch einschalten neuer Proglottiden die Zahl der Glieder an einem Bandwurme vermehrt wird." Again, under the section of the Embryology entitled "Die Uebertragung der Finnen in den Endwirth," etc. (p. 1592 on), he refers in a number of places to the appearance of the segments, but makes only indirect mention of the order in which they appear, the evident reason for this being that there is only one method of proglottid formation known.

On page 1597, Kuchenmeister, von Siebold, Lewald, Haubner and Leuckart are quoted as having studied this stage of the development in a number of forms about 1850, since which very little attention has been paid to it. Leuckart's first work upon this point appeared in 1856 and was subsequently elaborated in the editions of his text-book.

On p. 1600, Braun says: "Die näheren Vorgänge bei der Bildung der Proglottiden sind uns bisher noch ganz unbekannt, nur die Anlage und Entwickelung der Genitalien ist bei manchen Arten erforscht."

From indirect references at other places in the volume it is clear that the accounts current in our text-books are justified by the absence of any published description contradicting the accepted method of proglottid formation.

The amount of literature published on Cestodes up to 1896 is enormous, but the manner of proglottid formation was established and had been explicitly described for so long a time before this date that one can hardly believe any marked exception to the universal method could have escaped the notice and comment of so thorough a reviewer as the author of this monograph has shown himself to be. Upon such a point as this I feel that Braun may be taken as reliable and that I am justified in the conclusion that no such process of proglottid formation was described previous to 1896.

I have consulted a considerable amount of the literature on Cestoda appearing since that date, at first hand, and have cov ered the abstracts and titles of cestode papers as they appear in the *Zoölogische Jahresbericht*, without finding anything to indicate that accounts have been published of a method of proglottid formation other than the one commonly accepted as the universal.

Papers upon other phases of the proglottid question have appeared, as for example one by Lühe, '98, who, in his examination of the segmentation of *Ligula*, found that the segments which occur at the anterior end do not correspond to the arrangement of the genitalia where these occur toward the posterior end of the segmented area and hence, that this case is not one of true proglottid formation but rather to be considered as a differentiation of another sort. While there may be some homology

between these pseudo-proglottids of *Ligula* and the "anterior" proglottids of *C. laciniatum*, I have made it clear that the greater part, if not all, of these structures appearing at the anterior end become true proglottids in the form I studied.

Instances of irregularities like the occurrence of half proglottids are noted by Braun, p. 1225, and dismissed as abnormalities. Such a condition has recently been discussed by Child ('02) for *Moniezia*, but this again has nothing to do with the points I have brought out for *C. laciniatum*. The fact that no exception to the typical method seems to have been recorded is to my mind justification for an extensive account of the mode of proglottid formation which I have observed in *C. laciniatum* and such an account is therefore given in the section which follows.

With the exception of the work by Linton, who described the species, I know of nothing which has been published upon *C. laciniatum*. This author in his original description (Linton, '86, p. 474, Pl. III.) speaks of small specimens and of the segments occurring at either end in somewhat larger ones. The posterior segments are, he says, "totally unlike those of the adult" and "are evidently evanescent." It is, therefore, likely that Linton had among his specimens the various stages which I have described, but he paid little attention to them and hence his brief paragraph does not bring out their significance.

The "long-necked" specimens were also observed by Linton and are referred to on several occasions. In his "Report of Parasites Collected in 1898" (Linton, '99) he records several instances, and in an earlier paper ('89, on pp. 800–801) and Pl. VII., Fig. 4, he describes the "long-necked" forms and others with a reduced number of segments in such a way as to make it clear that he has seen all the stages up to the point where the segmentation of the neck region begins. While not thinking that they justify the erection of a new species he speaks of them as a variety (longicolle) of *C. laciniatum* and suggests that they may represent the transition to another species. It is thus evident that Linton being interested primarily in the systematic work, did not examine this point sufficiently to ascertain the real significance of such stages. The importance of his observations for my work is that they confirm my statement that a certain pro-

portion of long-necked specimens are always found when any large number of the individuals of this species is examined. This is borne out by the observations of some of my colleagues on the staff of instruction at the Marine Biological Laboratory, who have frequently observed such individuals among the specimens used by the students. In this connection it may be said that all the specimens which have been examined by these men, by Linton and by myself have been taken between June 15 and September 15. Since the sand sharks are not present about Woods Hole from November to June (Smith, '97, p. 89) one could not obtain data throughout the year. It would, however, be desirable to examine the worms from sharks taken as late and as early as possible, for there may be seasonal relations such as obtain in the period of maturity in other forms,[1] and one might find at another period a relatively larger number of "long-necked" forms.

III. Detailed Account of the Proglottid Formation.

(*a*) *Material and Technique.* — The *C. laciniatum* from which the data here presented have been obtained were collected at Woods Hole, Massachusetts, during several summers (1902–'04). I have examined a large number of specimens which have accumulated since I began work upon this form and by selecting out the different stages have been able to demonstrate every step in the history of the strobilla with an abundance of material. In some cases, the specimens were killed under the pressure of a cover slip, but for the most part they were taken from lots which had been killed by placing for a moment in fresh water and immediately transferring to the killing fluid. All the material used in this study was fixed in saturated sublimate with 5 per cent. acetic. Almost any good stain will bring out the division lines between proglottids, but for determining the location of the reproductive organs in whole mounts Partsch's alum cochineal gave most excellent results.

(*b*) *Proglottid Formation in the Larval Worms.* — Out of an abundance of material, I have been able to demonstrate again and again that, in the case of young worms which have presum-

[1] See the remarks of Braun, p. 1461, upon the "time of reproduction," etc.

ably just entered the shark, the proglottids originate as follows: The young worm which is recognizable by the small size and the proportions of its scolex region (Fig. 1, Pl. IV.) at first forms segments from behind forward in the typical cestode fashion (Fig. 2, Pl. IV.). The youngest specimen I have found (Fig. 1, Pl. IV.) appears to have something missing from the posterior end, but a comparison with Fig. 2 which is a little older would indicate that not more than one proglottid has been lost. The progress of this early segmentation, the increase in size and the changes in the proportions of the scolex parts are seen by comparing Figs. 1, 2 and 5, which are all drawn on the same scale. The terminal proglottid in such stages is invariably long as in Figs. 2 and 4, but in older specimens (Fig. 7) which show no signs of having lost any segments at the posterior end one is very likely to find it much shorter. In Fig. 3, for example, there is a very long terminal proglottid which is bent twice in places which are clearly not lines of division between segments. Figs. 4 and 6 represent specimens in which the most posterior line of division was much fainter than the lines anterior to it. This suggested the possibility that the last two proglottids in such cases had originated by the division of an earlier single terminal proglottid. I have often found specimens which suggested very strongly that such a division of the earlier proglottids had taken place, but since it is well-nigh impossible to make certain of such a process when specimens can only be taken and arranged in series after being studied, I do not wish to say more regarding this than that such cases as shown above have frequently come to my attention and suggested the possibility of the subsequent division of the elongated segments found at the posterior end in very young worms. The formation of simple rectangular segments from behind forward progresses steadily (Figs. 6 and 7) until from 40 to 60 of these "posterior" proglottids have been developed. There then begin to appear in the region just behind the scolex other proglottids. These "anterior" proglottids are from the first different in shape from the ones at the posterior region and this difference is, as will be shown, continued into the adult. The most important fact about these "anterior" proglottids is that they are differentiated from in front backward. Fig. 10 shows a specimen in which a number

of these "anterior" proglottids have appeared, Fig. 7 one in which there are a few more and the differences between the proglottid outlines is more apparent. These two figures moreover illustrate the absence of a very definite relation between the number of posterior segments formed and the time at which the anterior ones begin to appear, for in Fig. 7, there are 56 posterior and 10 anterior as against 66 posterior and in 6 anterior in Fig. 10. The marked differences in contour are better seen a little later, when there are more anterior segments present. Fig. 8 illustrates this and shows that the oldest ones have already assumed the shape so very characteristic for the adult proglottids of this region, and even in those which are just appearing (Fig. 8) the suggestion of the four lappets which occur on the posterior margin can already be seen. The contour of segments from this region is seen again in the figures of Pl. VI. and a stage which is practically the adult is shown in Fig. 25, *a*, Pl. VII. The four lappets are very mobile in the living specimen and their appearance differs considerably according to their state of contraction and the angle from which the strobilla is viewed as will be apparent from a glance at Figs. 14, *a*, Pl. V., 16, *a*, Pl. VI. and 25, *a*, Pl. VII.

The "posterior" segments have in their final condition four flattened lappets (Figs. 14, *n*, Pl. V., and 25, *c*, Pl. VII.). This sculpturing is distinctly different from that of the "anterior" segments until the latter come to occupy the terminal position (Fig. 15, Pl. VI.), when they assume the contours of typical posterior proglottids.

The formation of the posterior segments continues rapidly — (Figs. 10, Pl. IV., and 12, Pl. V.) and this region presents an appearance identical with that ordinarily met with in the cestode strobilla. The anterior proglottids develop more slowly and their total number is always much less than the posterior ones, hence, the region where the two meet and where the non-segmented zone is finally obliterated is well towards the anterior end.

The total number of posterior segments can be determined only approximately because there is always the chance that some have been lost either under natural conditions or during the handling incident to collection and preservation. Taking the maximum

number in the longest specimens it appears that there may be formed upwards of 400.

In the case of the anterior proglottids the total number formed can be ascertained with more certainty for it is only necessary to count them in specimens where the zone is just being obliterated. Such counts show that there may be as many as 604 proglottids formed in the anterior region. I have found a few cases in which the zone seemed about to be obliterated when there were only 30–35 anterior segments, but in the great majority of such cases their number runs well up toward 60 and I think 50 + would be a fair statement of the average number.

The transition of the posterior segments into the unsegmented zone is always gradual, as will be seen in Figs. 13, Pl. V., 23 and 25, *b*, Pl. VII. On the other hand the anterior segments may show a gradual transition into the zone (Fig. 13, Pl. V.) or the transition may be more or less abrupt (Figs. 23 and 25, *b*, Pl. VII.) The condition shown by Fig. 25, *b*, is the more common one for the anterior proglottids while that shown in Fig. 13, Pl. V., is typical for the transition of the posterior ones into the zone.

When the zone is finally obliterated it is no longer possible to tell where it was located for the proglottids of the anterior end, with their four-pointed lappets, gradually change into the straight transverse lines which separate the many immature proglottids in the middle region of the chain. The adult condition thus obtained is represented by Figs. 14, *a–n*, in which typical regions of the body have been drawn and the length of the chain between such regions shown by straight lines. The peculiar anterior proglottids (Fig. 14, *a*) still show their characteristic outlines as far back as the 110th proglottid from the scolex (Fig. 14, *c*) from which we may conclude either that the specimen figured had formed a greater number of anterior proglottids than I estimated as the average, or that some of the proglottids formed behind the non-segmented zone have assumed the character of immature anterior proglottids. From the condition shown at the 110th segment of Fig. 14, *c*, we reach after a distance represented by the line *d* a region of very close-set immature proglottids which are separated by straight transverse lines without any sculpturing

of the margins. This condition is continued for a distance shown by the line *f*, before the rudiments of the reproductive organs are clearly discernible. When a place is reached where these can be definitely made out (Fig. 14, *g*) it is seen that the proglottids are beginning to assume the outlines characteristic for the posterior members of the chain. In *g* of Fig. 14, we find the reproductive organs well marked out at the region just in front of the 386th proglottid. They are first apparent in the whole mount at about the region of the 325th proglottid. From the region shown by *g* of Fig. 14 to the posterior end the transition along *h* and *i* to the 471st at *j* is gradual and from this latter point through *k*, *l* and *m*, to the terminal region, shown by *n* of Fig. 14, the change consists more in the elongation of the proglottid than in its growth in bulk. The 506th proglottid which terminates the chain shows fully developed reproductive organs and some eggs accumulated in the uterus and is farther advanced than many of the free motile proglottids one finds in the intestine.

(*c*) *The Reduction of the Primary Strobilla and the Formation of the "Long-necked" Stage.* — Such a specimen as the one just described may be termed *a young adult*, for it still has about the number of proglottids which we have reason to believe is not far below the maximum and it is already liberating ripe segments from the posterior end. The following points are of importance in this connection: First, the reproductive organs do not begin until well back of the 300th proglottid. Though sections might show them being laid down in front of this region, there is in any case a long region extending beyond the anterior half of the worm in which they have not yet appeared. Second, the non-segmented zone has been obliterated and the place where it disappeared can no longer be recognized. Third, there is no point in the chain where there is anything to suggest the interpolation of new segments. I have examined a large number of specimens in this and in the stages next succeeding without finding the slightest indication that new proglottids are added in the region back of the solex or elsewhere.

The only change which takes place in such a specimen is the continual dropping off of ripe proglottids from the posterior end and the steady advance toward the scolex of the region of ap-

pearing reproductive organs. Specimens are found in all stages intermediate between Fig. 14 and those which like Fig. 16 have a much smaller number of segments. It thus seems clear that such a specimen as is shown by Fig. 16, *a*, *b*, is to be regarded as derived from the "young adult condition" of Fig. 14 by the progressive loss of ripe proglottids and the advance anteriorly of the differentiating reproductive organs. In the specimen just cited (Fig. 16, *a*, *b*) there is a total of only 96 proglottids in the chain and the terminal ones are ready to be set free. The reproductive organs which in the whole mount are recognizable in the 26th proglottid have advanced beyond the point (30th) which we have fixed as the very minimum for the number of "anterior proglottids." There is as yet no sign of the appearance of new proglottids to make good those which have been shed. We might refer to this as an adult which is approaching the old age period of the strobilla in contrast to such a "young adult" as Fig. 14 shows.

Specimens with a smaller number of proglottids and showing the posterior ones well matured have been frequently observed, but when the number has been reduced much beyond this point (Fig. 16) one finds an important change occurring in the region between the first of the old "anterior proglottids" and the scolex. This change consists in the growth of this region into what I shall call a "neck" which soon separates the scolex and the first proglottid by a very appreciable distance (Fig. 21, Pl. VII.). In this figure the reproductive organs are recognizable in the 17th proglottid and probably occurred further forward but the specimen from which this was drawn was poorly stained for this point. Such a specimen as Fig. 15, Pl. VI., shows the reproductive organs well established in the 5th segment while in the 10th and last one they are fully matured. Such a case as Fig. 18, Pl. VI., has clearly come from an old chain which has formed a "neck" to which two of the old anterior proglottids are attached. It is very possible, however, that this specimen which I found among many other preserved specimens has had some segments detached in the collection or subsequent handling.

Specimens in which the neck has become well established next show a segmentation at the posterior end of the neck region (Fig.

17, VI., and 22, VII.), and this segmentation progresses from behind forward as did the formation of "posterior segments" previously described. When from six to twelve such segments have been established there begins a segmentation of the front end of the neck. This proceeds from in front backwards as did the formation of "anterior" proglottids at an earlier stage. Specimens showing the above changes are shown in Figs. 17, Pl. VI., 22, Pl. VII., 20, Pl. VI., and 24, Pl. VII. The number of proglottids of the old chain which are present when the neck begins to appear and later the number present when the segmentation of the neck begins, appears subject to a considerable variation. For example, Fig. 15, Pl. VI., and Fig. 21, Pl. VII., are specimens which when the state of contraction in each case is considered appear to have the neck developed to about the same extent, but the number of remaining proglottids is ten in one case and thirty-four in the other. Nothing can be determined on this point from Figs. 17 and 18, Pl. VI., or 22 and 24 of Plate VII., for although Figs. 18, 22 and 24 had the number of proglottids indicated when they were taken from the shark they may have been mutilated when the valve was cut open, while in the case of Fig. 17 I have no record of the condition when collected and hence suspect the possibility that some segments may have been detached by accident. Again, in Fig. 20, Pl. VI., is shown a specimen in which the neck has both anterior and posterior segments though forty-nine of the old proglottids remain. This is the maximum number of the old segments of which I have ever found upon a necked specimen. It would seem, therefore, that the neck region appears when the supply of proglottids comprising the earlier chain has become so much reduced that the developing reproductive organs begin to appear well into the region occupied by proglottids which must have had an "anterior" origin in the embryo, but the number of primary proglottids remaining when the neck appears is quite inconstant.

In the early part of my work, when I had not obtained so complete a record of the conditions in the "necked" specimens, I was inclined to question whether the segments formed at the anterior end in the larval stage (Fig. 8, Pl. IV.) were to be regarded as proglottids in the same sense as those developed posteriorly,

i. e., whether we had not to do with what one might term pseudo-proglottids as distinguished from the true or posterior proglottids. This led me to consider the criterion by which one would determine whether a region of the body was a proglottid or not. My view is that we are justified in applying the name proglottid to any region of the body which is set off from the neighboring parts by constrictions and which contains a complete set of reproductive organs. From many specimens which were examined when the non-segmented zone was almost obliterated, I found as previously stated that no specimens observed showed less than 30 + segments of "anterior" origin and that the number produced in this region was 50 + in the majority of cases. If this is so, specimens like Figs. 15, Pl. VI., and 21, Pl. VII., show us that such "anterior" proglottids may eventually become sexually mature and be shed off the same as any other in the chain. Moreover, as will be seen in the two figures just cited, anterior proglottids thus becoming ripe change their shape by the flattening of the four-pointed lappets of the earlier stage and assume exactly the shape characteristic for a ripe proglottid of posterior origin. I think, therefore, that we may dispose of any question as to whether the segments originating from in front backwards are to be considered as true proglottids.

The new segmentation appearing at either end of the neck region is identical with that which occurs in the young worm save that the "anterior" proglottids begin to appear before so many of the posterior ones have been developed. Were it not for the single terminal segment and for the slightly greater size of the bothria on the scolex one would certainly class specimens like Figs. 22 and 24, Pl. VII., as young worms similar to those shown in Figs. 7 and 10, Pl. IV., and 11, of Pl. V. The size of the bothria would not be very reliable in preserved specimens, but the single terminal proglottid with its four-pointed lappets would seem unquestionably to be the first anterior proglottid of a preceding strobilla.

I have not been able to secure stages showing that the segmentation of the neck region finally results in a new strobilla. The difficulty in distinguishing specimens, which have passed farther along the way to this condition than Fig. 24, from stages in

the primary strobilla would be considerable unless one of the old anterior proglottids still remained and I have not felt that the prospect of finding such stages justified my delaying the completion of this paper until I had another opportunity to examine a large amount of fresh material in search of them. This segmentation of the neck region is either the beginning of a new strobilla which to distinguish from the earlier we might call the secondary strobilla, or it is simply an abortive attempt in the old age of the individual and comes to naught. From what we know of the longevity and continued production of proglottids in a few other cestodes, Braun, p. 1604, I think the presumption is distinctly in favor of the former view, *viz.*, that the segmentation which I have observed beginning in the neck region is the first step in the formation of a new strobilla. Though it would be almost impossible to demonstrate the formation of other strobillæ from new neck regions after the secondary one has in turn become exhausted, we may, I think, consider such a process as not at all unlikely. Hence, we have the suggestion that what may be called primary, secondary and tertiary, etc., strobillæ are successfully headed off by a single individual scolex.

(*d*) *An Exceptional Condition, Perhaps to be Explained as Regeneration after Chance Mutilation.* — Two specimens have come under my observation which apparently do not find any place in the above scheme of development. One of these is shown in Fig. 19 of Pl. VI. In this we have a body with a definite zone region and the anterior and posterior segments which characterize a worm developing the primary strobilla. In contradiction to these features, we find a well-developed neck region. The size of the bothria gives no trustworthy evidence of the age in these specimens. I think these two exceptions may be explained as cases of regeneration after a chance mutilation occurring under natural conditions.

It will be noted if Fig. 19, Pl. VI., is compared with Fig. 12 of Pl. V., that the number and development of the anterior proglottids is relatively much greater in Fig. 19 than we find in the normal development of the primary strobilla. If such a specimen as Fig. 11 or Fig. 12, Pl. V., had been cut in two somewhere near the forward region of the posterior proglottids and

by this loss of a considerable portion of the young strobilla, the part having the scolex had been stimulated to form a neck for the production of more segments, we should have such a condition as Fig. 19 shows. It is commonly assumed in the theories regarding the nature of the cestode body that the Cestoda have great powers of regeneration. For example, Lang ('88) used this supposed capacity for regeneration as the starting point in his explanation of how the strobillization was first introduced into cestode phylogeny. We have, however, little in the way of direct evidence either from observation of specimens regenerating under natural conditions or from accounts of their regeneration under experimental control. The difficulties of technique have so far as I know prevented any experimental confirmation of this supposed power of regeneration. Still the assumption seems reasonable and the difficulties could doubtless be overcome if it seemed worth while to determine what regenerative powers the Cestoda do actually possess. In the case of *C. laciniatum* one should be able to test the explanation I have just given for these exceptional cases by the mutilation of young specimens in the way indicated. If one could overcome the difficulties and keep the worms alive by introducing them into another shark or otherwise, I think some very interesting data might be obtained by the mutilation of strobillæ in different stages and a study of the regeneration if such a process occurs.

(*e*) *Histology of the Non-Segmented Zone in Young Worms and of the Neck Region in Older Specimens.* — I have examined sections of this region which is the place where differentiation appears to be most active hoping to find some characteristic features by which its histology might be distinguished from that of the ordinary proglottid, but with the material available have not been able to recognize any such distinguishing features. If nuclear division is in progress it must be amitotic such as Child ('04) has recently described for *Moniezia*, for the material I have would show clearly the existence of mitotic figures. I have endeavored by staining after the method followed by Child to determine the presence or absence of such division but the fixation is not quite good enough for this point. This histology seems to be much the same as that of the formed proglottid save that

the muscle fibers are less developed. In the young worm the zone has a circular outline in section while in the neck region it is somewhat flattened. Fig. 26 which represents the structures found in this zone of a young worm shows that there occur only the structures which are regarded as typical for the adult cestode (Schneider, '02, p. 311, Fig. 324, and Braun, Pl. XLVII.). There are circular and longitudinal muscle fibers between the cuticle and the palisade of subcuticular cells, the latter being grouped so that they form lines between the outer portions of these cells. The parenchyma contains scattered nuclei, some with cytoplasm aggregated about them as in a definite cell body. One member of each pair of water tubes appears in the figure. The results of this examination are disappointing, for unless there is an active amitotic division, which I fail to make out with this material, there is nothing which would suggest the active differentiation which is taking place. However, I do not offer this as in any way an adequate discussion of the histology of this region. Such an account I hope to give at some future time when I have proper material for a thorough examination. This is merely a statement of what I have been able to accomplish along this line thus far.

IV. General Considerations.

If my interpretation of the stages found is correct, we have in *Crossobothrium laciniatum* a method of proglottid formation which is radically different from the one now accepted as universal. Also, there seems to be more than one strobilla formed so that we may speak of primary and secondary strobillæ and perhaps there are even more. This is in marked contrast with such forms as the tænias which present a continuous growth and differentiation in the neck region to make good a continuous shedding of ripe proglottids. If we try to analyze the proglottid formation here described and to compare it with the process which occurs in other cestodes, we may do so in the following way. The appearance of the first proglottids at the posterior end is homologous to the method now accepted as the universal one. Such a stage as is shown by Fig. 5, Pl. IV., and from this on to the time when the "anterior" proglottids begin to appear, may

be regarded as a cestode with an unusually long neck region. The essential point of difference comes when proglottids begin to differentiate at the anterior end of this neck *in the reverse direction.* It must however be noted that subsequently the front part of this neck region becomes the neck of the next strobilla which at its posterior end differentiates from behind forward. It is thus possible to regard the segment formation of *C. laciniatum* as derived from the ordinary type by the acquisition of a long neck and subsequently its differentiation in the reverse order at the front end.

While it is clear that the proglottids of the adult are serially homologous throughout the worm, we may ask ourselves whether there can be found any good reason for the modification of structure which is so noticeable in those of the anterior region. If we consider the case of such a "young adult" specimen as that shown by Fig. 14, *a–n*, Pl. V., it is clear that this modification of the four posterior corners into sharp projections is not a characteristic belonging exclusively to the proglottids which had an anterior origin. In this figure if we examine *a*, *b* and *c* we find that the area occupied by proglottids having the shape in question extends considerably beyond the region occupied by the proglottids which probably had an anterior origin. In such a specimen we can no longer determine where the non-segmented zone finally disappeared, but as has been pointed out there are never many more than 60 anterior ones and hence the zone must have been not far from the 60th segment. The proglottids having the shape in question are found extending even back of the 110th as shown in *c* of Fig. 14. This means when applied to such a figure as 13 of Pl. V., or 23 of Pl. VII., that some of the forward members among the simple proglottids behind the "zone" are later made over into the anterior type, and in Fig. 25, *b*, Pl. VII., we see that this may begin even before the zone is obliterated. It is thus clear that the fact of a proglottid having an "anterior" origin is not the factor which has determined the peculiar shape of the segments at the front end of an adult *C. laciniatum*, but that the antero-posterior differentiation which exists extends beyond the region of the obliterated "zone" and into that of the posterior segments. The cause would then seem

to be something which affected the anterior end of the strobilla independently of the origin of the proglottids involved.

It has occurred to me in collecting the worms from the spiral valve that perhaps the four projections which characterized these forward segments of the adult strobilla, may be regarded as an adaptation by which the worm is better able to retain its hold with the scolex. The passage along between the folds of the semi-solid feces which are often found in the valve, must often draw out the bodies of these cestodes and tax the hold of the scolex to a corresponding degree. The projections in question would be very effective in preventing the front part of the worm from slipping backward and would thus relieve the strain on the scolex. I believe that we must look to something in the conditions which act upon the anterior region of the strobilla and not to the anterior or posterior origin of any given proglottid, to account for this feature of *C. laciniatum* and the above suggestion is given as a possible explanation.

Since the theories regarding the nature of the cestode body have taken into consideration the supposed universal method of proglottid formation we may consider what bearing the facts here established have upon such theories. At the outset I will say that I do not think this single case of a small number of true proglottids developed anteriorly and in the reverse direction will justify any sweeping modification of existing views, for it is only fair to suppose that the posterior proglottids of *C. laciniatum* are strictly homologous to the entire strobilla in other cestodes and we may regard *C. laciniatum* as a species in which another method of proglottid formation has been superposed on an older and more universal one. The history of our theories regarding the nature of the cestode body is extensively summarized by Braun in the Cestode volume of Bronn's Thierreich. Beginning with the earlier conception of the chain as composed of proglottids which had become united, he traces the history of the theory developed by J. P. van Beneden, Siebold, Leuckart and others, which viewed the Cestode at a "polyzoötic" organism, *i. e.*, that despite the physiological unity of the chain of proglottids we are dealing, from the morphological standpoint, not with a unity but with an animal stock. This view has been main-

tained by some to the present day, having been defended more recently by J. v. Kennel in his "Lehrbuch der Zoölogie." On the other hand it has been subject to considerable attack which is summarized in the theory accepted by Lang and Korschelt and Heider. The latter authors hold that the scolex and a single proglottid represent the individual and are equal to the body of a trematode and that the present organization of the cestode body has been attained by the reduplication of that region which contains the reproductive organs. In a way this opinion is not so different from the other view since we may think of this reduplication as a budding process in which only the posterior half of the individual is formed.

Broadly speaking, the discussion reduces itself to the question whether we shall consider the segmented condition of the cestode as having arisen by the loss of individuality and the adherence together of originally complete individuals which were formed by budding, or as arisen by the elongation and reduplication of certain parts in a single individual. It is like the question of metamerism which though most commonly explained in accordance with the latter (Lang, '82, and others) has nevertheless been accounted for by some (Whitman, '99) along the line of the former view. Personally, I agree entirely with Braun in the concluding paragraph of the discussion above cited, that the theory one will incline to is largely a question of temperament since the phenomena can be interpreted in either way. The facts brought out for *C. laciniatum*, while they show a new method of proglottid formation do not turn the scale in favor of either interpretation. If such a condition as is here described could be shown to be more widespread in the cestoda and thus the one we now regard as primitive should lose some of its importance we might then believe that a segmentation proceeding from both ends toward the middle is better explained upon the hypothesis that reduplication of parts has occurred in a single individual than upon the theory that it has arisen through a process of strobillization identical with that by which the ephyræ of a scyphozoan are formed. But the facts regarding *C. laciniatum* stand alone and we can at present claim no more than that the formation of proglottids in the cestoda deserves renewed investi-

gation particularly in any forms which suggest a departure from the type of development hitherto supposed to be without exception.

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COLUMBIA, MO., June 15, 1906.

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PLATE IV.

Figures showing stages in the proglottid formation of *C. laciniatum* as found in the spiral valve of the shark, *Carcharias littoralis*. All from camera lucida outlines. The magnification is given for the figures as reproduced.

FIG. 1. The youngest specimen obtained. The outline at the posterior end indicates that at least *one* proglottid has been lost, but the small size of the scolex shows that this one is slightly younger than the complete specimen shown in Fig. 2 and hence it is unlikely that more than one proglottid is missing. × about 50 diameters.

FIG. 2. A complete specimen slightly older than that of Fig. 1. The first proglottids are forming from behind forwards in typical cestode fashion. × about 50 diameters.

FIG. 3. The posterior region of a specimen in about the stage of Fig. 2, showing a very long terminal proglottid which is bent in two places, but does not show any lines of division. Compressed when killed and hence apparently wider than Fig. 2. × 18 diameters.

FIG. 4. A similar specimen showing a condition which suggests that a long terminal proglottid of an earlier stage has begun to divide into two. × 18 diameters.

FIG. 5. A specimen slightly older than Fig. 2. The scale of 50 diameters being the same as that of Figs. 1 and 2 shows the marked increase in size. × 50 diameters.

FIG. 6. Posterior portion of a specimen somewhat older than Fig. 5. To show typical dimensions of proglottids at this stage. The last division line is faint as though a longer terminal proglottid had just divided. × 18 diameters.

FIG. 7. An entire specimen in which the "anterior proglottids" are appearing. This is actually much larger than the specimen shown in Fig. 5. × 18 diameters.

FIG. 8. The anterior segments of a specimen which still showed an extensive non-segmented zone. × 42 diameters.

FIG. 9. The last six segments from the posterior end of a specimen similar to the one used for Fig. 8. × 42 diameters.

FIG. 10. A specimen of about the same stage as Fig. 7, but with fewer "anterior" and more "posterior" proglottids. A deeper constriction is noticed between the eighth and ninth proglottids from the posterior end. × 18 diameters.

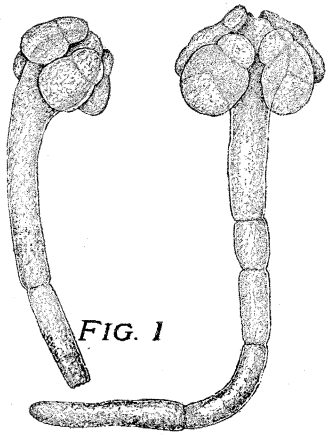

FIG. 1

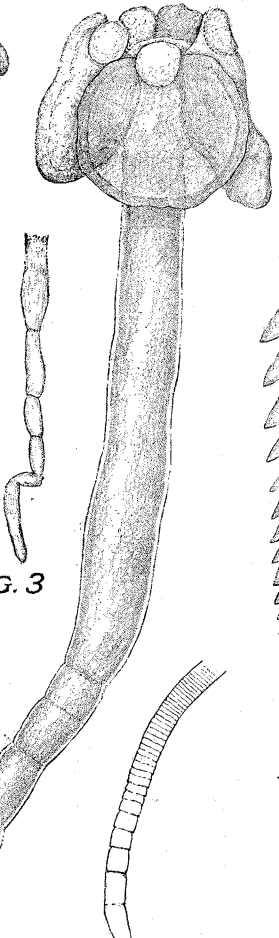

FIG. 2

FIG. 3

FIG. 4

FIG. 5

FIG. 6

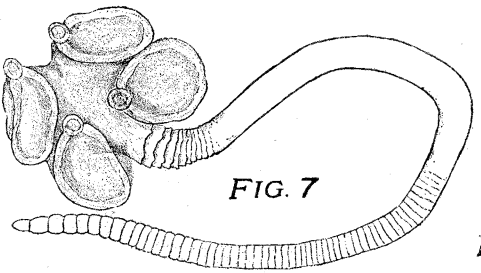

FIG. 7

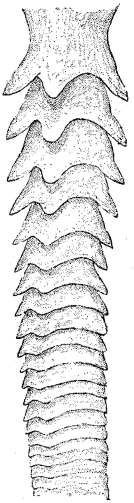

FIG. 8

FIG. 9

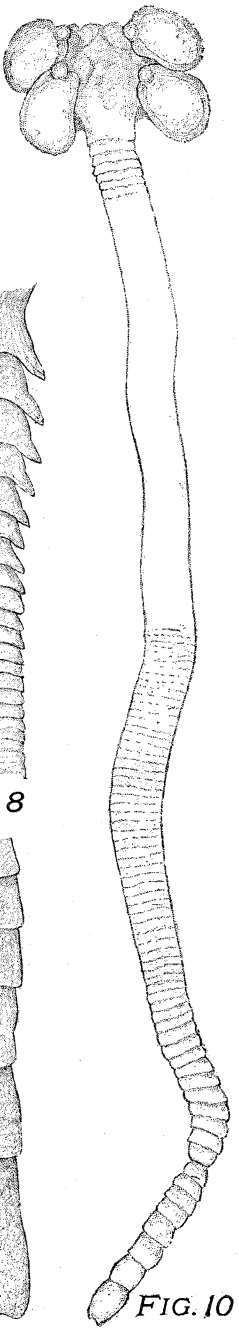

FIG. 10

PLATE V.

Figures of late stages in the proglottid formation of *C. laciniatum*. All from camera lucida drawings. The magnifications are for the figures as reproduced.

FIG. 11. A specimen with 8 "anterior" and 35 + "posterior" proglottids and showing a greatly elongated, non-segmented zone. × 12 diameters.

FIG. 12. An entire specimen in which the non-segmented zone has been greatly reduced by the appearance of 30 + "anterior" and 200 + "posterior" proglottids. × 12 diameters.

FIG. 13. The non-segmented region of a specimen in which the "anterior" and "posterior" segments are just meeting to obliterate the non-segmented area. Such a stage would be slightly beyond that of Fig. 12. In this instance there were 40 + "anterior," 250 + "posterior" proglottids. × 50 diameters.

FIG. 14, *a–n*, represents the appearance of the proglottids and their proportions in a young adult specimen which contains about its maximum number and is just beginning to liberate ripe proglottids at its posterior end. The length of the regions not drawn in is represented by the straight lines. The letters show the sequence in which the pieces should be arranged. The number of proglottids up to that point is marked at the posterior end of each section drawn. The total number of proglottids was about 506. × 12 diameters.

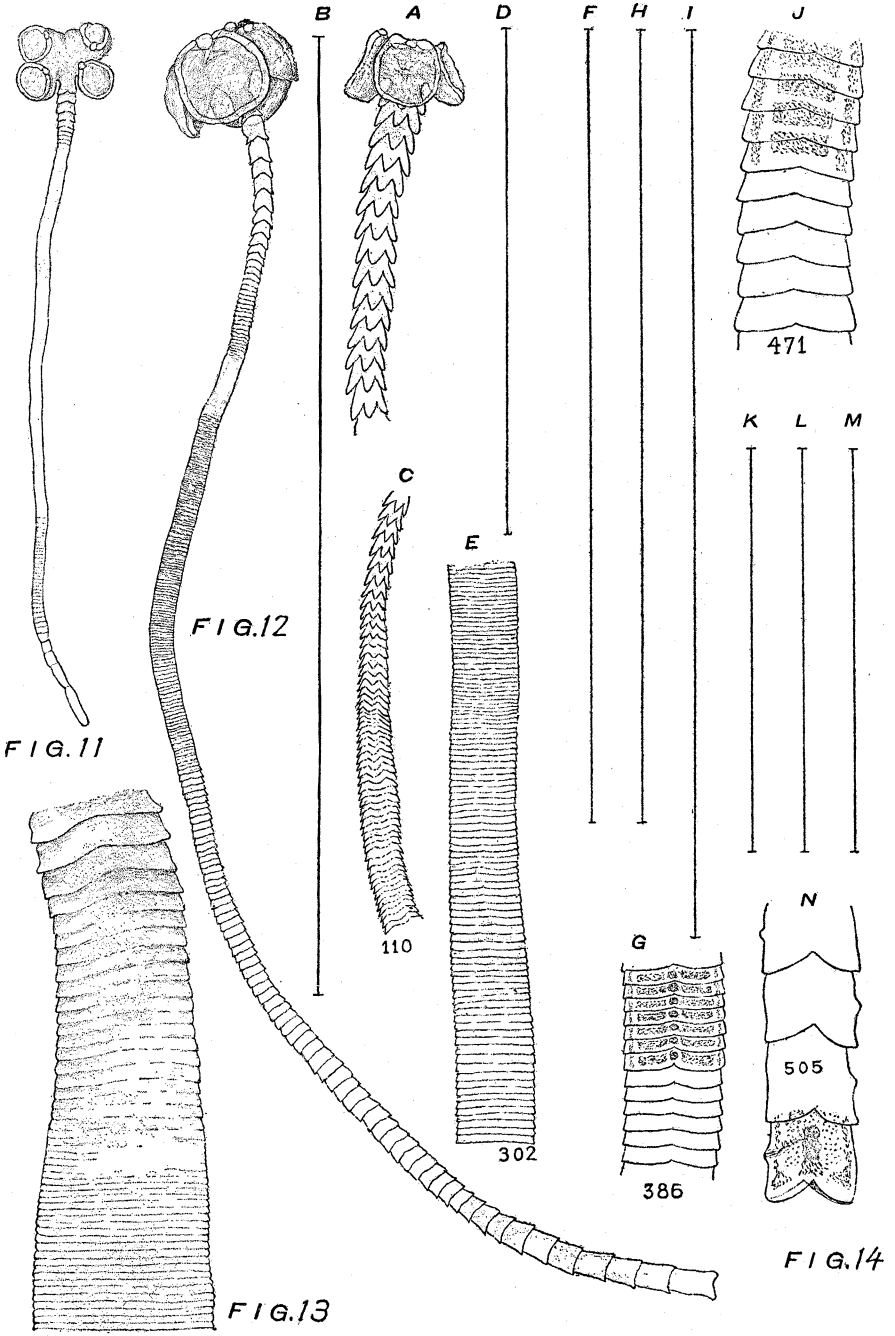

FIG. 11

FIG. 12

FIG. 13

FIG. 14

PLATE VI.

Figures showing the development of the "long-necked" specimens of *C. laciniatum*. All from camera lucida outlines. The magnification is given for the figures as reproduced.

FIG. 15. A "long-necked" specimen in which the reproductive organs can be made out as far forward as the fifth proglottid. The tenth and last is about ready to be shed. × 12 diameters.

FIG. 16, *a* and *b*. The entire length of a specimen which is approaching the "long-necked" condition. There is a total of 98 segments and the reproductive organs can be distinguished as far forward as the twenty-sixth proglottid. × 12 diameters.

FIG. 17. A "long-necked" specimen with 8 of the old anterior proglottids and the new segmentation begun in the posterior region of the neck. × 12 diameters.

FIG. 18. A "long-necked" specimen in which the new segmentation has not yet begun. × 12 diameters.

FIG. 19. A "long-necked" specimen with an unsegmented zone further back. See text for a possible explanation of this specimen. × 12 diameters.

FIG. 20. A "long-necked" specimen which has still 49 of the old proglottids in which reproductive organs can be made out as far forward as the third or fourth from the anterior end. The neck region has begun to segment at either end. × 12 diameters.

FIG. 15

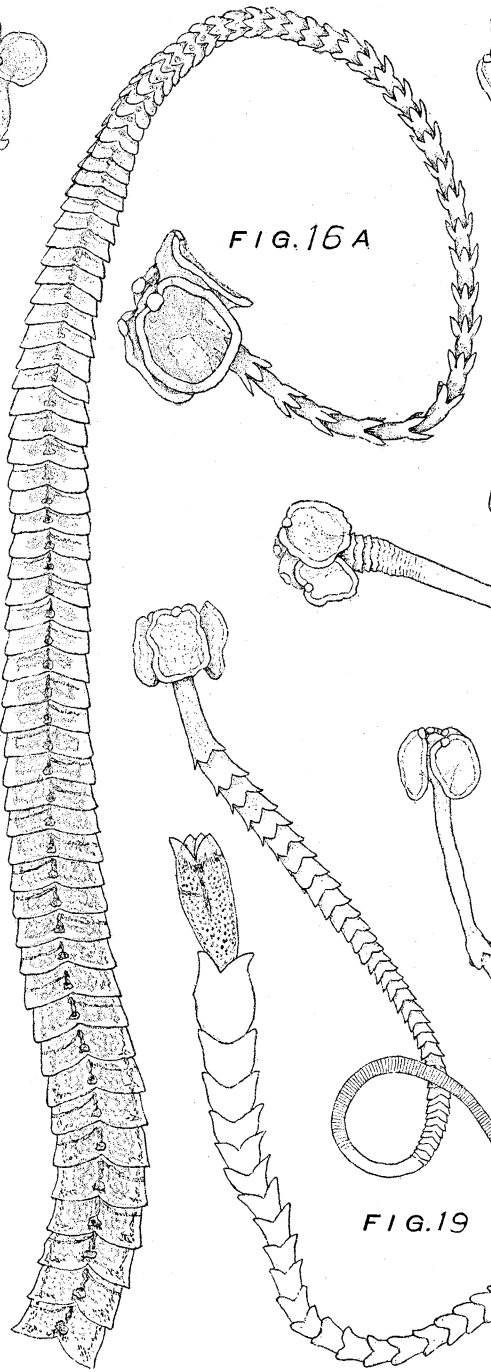

FIG. 16 A

FIG. 16 B

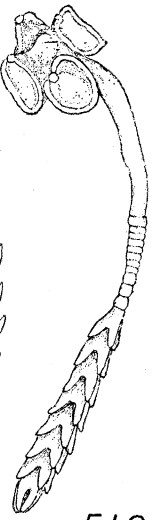

FIG. 17

FIG. 18

FIG. 19

FIG. 20

PLATE VII.

Figures showing miscellaneous points in the proglottid formation of *C. laciniatum*. All from camera lucida drawings. The magnification is given for the figures as reproduced.

FIG. 21. A specimen which has just entered the "long-necked" condition. There are 34 old proglottids. Reproductive organs probably occur in front of the seventeenth proglottid, but as the specimen was stained for another purpose this cannot be determined with certainty. × 12 diameters.

FIG. 22. A "long-necked" specimen with a single one of the old "anterior" proglottids attached and segmentation beginning in the posterior part of neck region. × 12 diameters.

FIG. 23. Part of a young specimen showing a short non-segmented zone with an abrupt transition of the "anterior" proglottids into the zone. There were in this instance 30 "anterior" and upwards of 217 "posterior" proglottids. × 50 diameters.

FIG. 24. A "long-necked" specimen with a single "anterior" proglottid of the earlier chain still attached at the posterior end and the new segmentation well begun at either end of the neck region. × 12 diameters.

FIG. 25, *a*, *b* and *c*. To show the characteristic shapes of the "anterior" (*a*) and the "posterior" proglottids (*c*). Also the unsegmented zone which separates them. All from the same specimen. There were 35 anterior and 250 + posterior segments. × 50 diameters.

FIG. 26. Part of a transverse section through the non-segmented zone of a specimen about the stage of Fig. 7, Pl. I. See discussion on page 216 of the text.

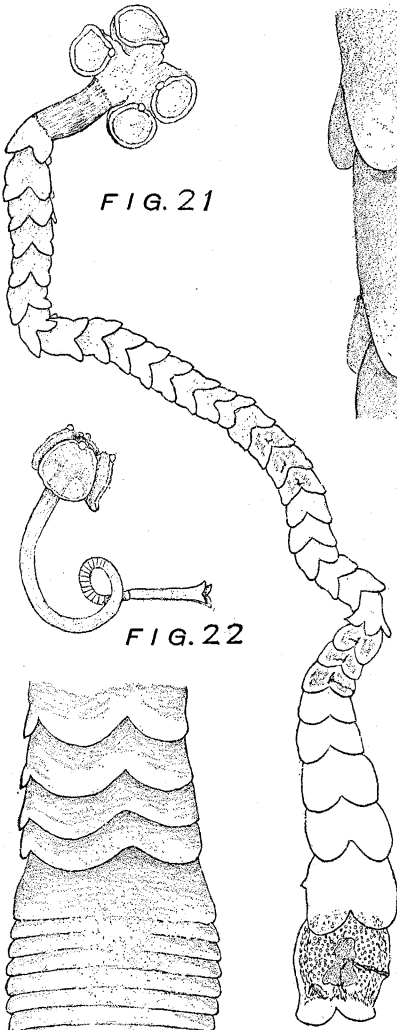

FIG. 21

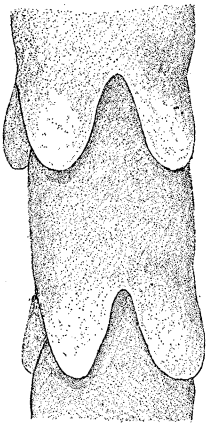

FIG. 25 A

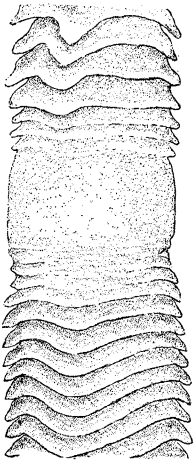

FIG. 25 B

FIG. 22

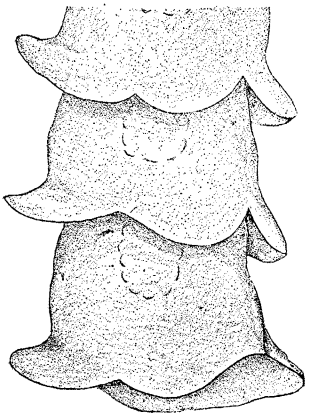

FIG. 25 C

FIG. 23

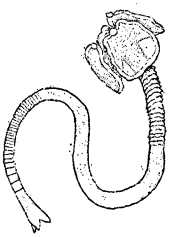

FIG. 24

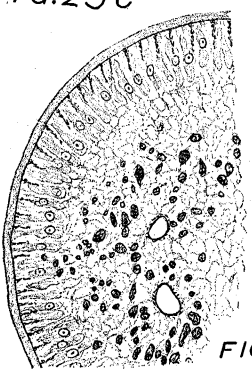

FIG. 26